

# Impact of Selected Electromagnetic Fields on Bone Turnover Markers in Rats

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**Abstract**—In this study the impact of electromagnetic field generated by mobile phone ( $f=900$  MHz), electromagnetic field with industrial frequency generated by high voltage alternating current transmission lines ( $f=50$  Hz,  $E=10$  kV/m), and also simultaneous action of those fields on the intensity of bone turnover process in male rats was estimated by means of analysis of serum concentration of calcium, phosphorus, as well as specific markers of bone formation and bone resorption. It was concluded that 4-week lasting exposure to both forms of electromagnetic field causes the intensification of bone turnover, resulting in the increase of serum concentrations of osteocalcin, pyridinoline and cross-linked alpha-2 N-terminal telopeptide of collagen type I. The obtained effect was dependent on frequency and power density of used electromagnetic fields.

**Keywords**—electromagnetic field; mobile phone; industrial frequency electric current; bone turnover; markers of bone formation and bone resorption; rats

## I. INTRODUCTION

In recent years in many experiments on animal models [1], [2], [3], [4], [5] and in few clinical trials [6], [7] it was found that electromagnetic fields with various values of frequency and power density influence the homeostasis of bone tissue. The most of previous papers in this field concern the effect of ELF electromagnetic fields with frequencies in the range of 5-50 Hz, that cause mainly stimulation and intensification of osteogenesis, resulting in acceleration of bone fracture repair, inhibition of bone tissue loss and its demineralization in osteoporosis patients, as well as increase of the contents of collagen fibers in the bone matrix [8]. Recently some papers were published, proving that electromagnetic fields with definite frequencies could modify the process of bone remodelling, especially in young individuals, by direct inhibition of process of osteoblastogenesis, as well as by stimulation of osteolysis.

## II. AIM OF STUDY

The aim of the study was to estimate the impact of electromagnetic field generated by mobile phones ( $f=900$  MHz), electromagnetic field with industrial frequency generated by high voltage electric current transmission lines ( $f=50$  Hz,  $E=10$  kV/m), and also simultaneous action of those fields, on the intensity of bone turnover process in male rats, by means of analysis of serum concentrations of calcium and phosphorus, as well as specific markers of bone formation and bone resorption.

## III. MATERIAL AND METHODS

### A. Experimental Animals

The experiment was performed on 40 male Wistar rats, in mean age of 10 weeks with mean initial body mass of  $180\pm 7,5$  g before the beginning of the experiment. In order to estimate the impact of electromagnetic field with frequency of 50 Hz generated between two electrodes of experimental system supplied with an alternating current, as well as electromagnetic field with frequency of 900 MHz generated by mobile phone (model Nokia 5110) the rats were divided into 4 equal groups (consisting of 10 animals) subjected to long-term exposure to electromagnetic fields with different physical parameters and different procedure of exposure or to sham-exposure.

### B. Procedure of Exposure to Electromagnetic Fields

Rats from examined group  $B_{1(s)}$  were exposed to electromagnetic field with physical parameters generated by typical high voltage alternating current transmission lines ( $f=50$  Hz,  $E=10$  kV/m), 22 hours a day (with a break between  $8^{00}$  and  $10^{00}$ ) for 28 succeeding days. Rats from examined group  $B_{2(s+m)}$  were exposed to electromagnetic field with identical parameters as in previous group ( $f=50$  Hz,  $E=10$  kV/m), that was also generated 22 hours a day for 28 succeeding days, and additionally during whole period of exposure cycle (28 days), every  $\frac{1}{2}$  hour by 8 hours daily, a mobile phone Nokia 5110 working in frequency range  $f=900$  MHz, placed under a cage with animals, was turned on and emitted a signal for 15 s. The mean value of power density of the electromagnetic field  $E_1$  registered during initializing of connection was  $85,3 \mu\text{W}/\text{m}^2$ , while the mean value of power density of the electromagnetic field  $E_2$  registered during lasting connection was  $17,0 \mu\text{W}/\text{m}^2$ . Rats from examined group  $B_{3(m)}$  were exposed for 28 succeeding days solely to electromagnetic field with frequency of 900 MHz generated by mobile phone, that was turned on similarly as in group  $B_{2(s+m)}$  every  $\frac{1}{2}$  hour by 8 hours daily and emitted for 15 s signal with physical parameters identical as in previous group. Rats from control group were exposed for 28 succeeding days to sham-exposed, during which they stayed in identical as examined animals environmental conditions, excluding the influence of electromagnetic field.

### C. Procedure of Laboratory Analyses

After 2-day period of adaptation in all rats a small specimens of blood (0,5 ml) was collected from the tail vein. The same procedure was repeated after the end of 1 and 3

week of exposure cycle. After decantation and centrifugation of the collected blood, a serum specimens were obtained, that were subsequently frozen in the temperature of  $-20^{\circ}\text{C}$ . After the end of the whole experiment the serum specimens were unfrozen in order to measure the concentrations of: marker of bone formation - osteocalcin (OC), markers of bone resorption - cross-linked alpha-2 N-terminal telopeptide of collagen type I (NTx) and pyridinoline (PYD), as well as total calcium and phosphorus [9], [10], [11]. After the end of a cycle of 28 daily exposures to electromagnetic field with physical parameters fixed for particular groups of exposed rats ( $B_{1(s)}$ ,  $B_{2(s+m)}$  and  $B_{3(m)}$ ) or 28 daily sham-exposures (control rats), animals were anaesthetized with use of a mixture of *xylazine* (10 mg/kg *ip*) and *ketamine* (100 mg/kg *ip*), and then specimens of blood (2 ml) were collected from the left heart ventricle. In obtained serum the concentrations of OC, NTx, PYD, as well as total calcium and phosphorus were measured. The analysis of calcium and phosphorus serum concentration was performed by means of colorimetric method with use of diagnostic kits Calcium-MTB and Phosphorus manufactured by Bio-Systems. In turn the serum concentrations of osteocalcin, cross-linked alpha-2 N-terminal telopeptide of collagen type I and pyridinoline were measured by means of colorimetric, immunoenzymatic method ELISA, with use of following diagnostic kits: Rat-MID Osteocalcin EIA (manufactured by Immunodiagnostic systems), Osteomark NTx Serum ELISA (manufactured by Osteomark) and MicroVue Serum PYD EIA Kit (manufactured by Quidel).

#### IV. RESULTS

In none group of rats exposed to electromagnetic field no significant changes of calcium serum concentration was observed, as compared to control group. The osteocalcin serum concentration in all groups of rats decreased in succeeding weeks of exposure cycle, in comparison to initial values. The serum concentrations of osteocalcin in rats from groups  $B_{1(s)}$  and  $B_{2(s+m)}$  were significantly higher, by 28,31% and 33,13%, respectively after first week of exposure cycle, by 21,63% and 8,48%, respectively after third week of exposure cycle, and by 66,05% and 21,87%, respectively after fourth week of exposure cycle, in comparison to control group. In rats from group  $B_{3(m)}$ , exposed to electromagnetic field generated by mobile phone a significantly higher serum concentration of osteocalcin by 27,39% in comparison with control group was observed, only after first week of exposure cycle. NTx serum concentration in group  $B_{3(m)}$  and control group decreased in 3 week of exposure cycle, while in groups  $B_{1(s)}$  and  $B_{2(s+m)}$  a distinct increase in the NTx serum concentration was observed, as compared to initial values. After 1 week of exposure cycle a significantly higher serum concentration of NTx in rats from group  $B_{3(m)}$ , by 13,00% in comparison with control group was observed, while after 3 and 4 week of exposure cycle a significantly higher concentrations of this marker were observed in all groups of rats exposed to electromagnetic field, after 3 week of exposure cycle they were higher by 38,43%, 43,41% and 15,65%, respectively and after 4 week of exposure by 24,29% in comparison with control group. In turn, the serum concentration of pyridinoline estimated after four weeks of a cycle of

exposure to electromagnetic field, in all groups of exposed rats ( $B_{1(s)}$ ,  $B_{2(s+m)}$  and  $B_{3(m)}$ ) was significantly higher, by 17,08%, 29,20% and 26,75%, respectively, in comparison with control group.

#### V. CONCLUSIONS

On the basis of the obtained results it was proved, that 4-week lasting exposure of rats to electromagnetic field with physical parameters generated by mobile phones and to electromagnetic field with physical parameters generated by high voltage alternating current transmission lines, as well as simultaneous exposure to both field causes intensification of bone turnover resulting in increase of serum concentration of osteocalcin (marker of bone formation) and serum concentration of pyridinoline and cross-linked alpha-2 N-terminal telopeptide of collagen type I (markers of bone resorption). It was also confirmed that the intensity of bone turnover under the influence of electromagnetic fields depends on the physical parameters of electromagnetic field as: frequency and power density.

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